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Original article

Evaluation of Napsin A and CK5/6 Expression in Non-Small Cell Lung Carcinoma (NSCLC): An Immunohistochemical Study

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Abstract

Background: Lung cancer is the most common type of cancer and the principal cause of death from cancer worldwide. **This study aimed to** evaluate the expression of Napsin A and CK5/6 in NSCLC, correlate immunohistochemical results of Napsin A and CK5/6 in differentiating between adenocarcinoma and SCC of the lung. assessment of the expression pattern of Napsin A and CK 5/6 as regards the available clinicopathological data available. **Methods:** This retrospective study was carried out on 40 lung cases; included 26 adenocarcinoma cases (Resection) and 14 cases of squamous cell carcinoma (Including 7 resection cases and 7 bronchoscopic biopsy cases). **Results**: Highly significant increase in CK5/6 score and expression, in SSC group; compared to ADC group ($p < 0.001$). ADC had a highly significant positive correlation with Napsin A score ($p < 0.01$). In the current study Pearson's correlation analysis shows that SSC, had a highly significant positive correlation with CK5/6 score. **Conclusion:** From our findings we can conclude that Napsin A score can predict patients with ADC from patients with SSC. the increase in SSC histology; had an independent effect on increasing the probability of CK5/6 expression. The decrease in LN; had an independent effect on increasing the probability of Napsin A expression.

Keywords: Napsin A; CK5/6; NSCLC; Immunohistochemical.

Introduction

Lung cancer is the most common type of cancer and the principal cause of death from cancer worldwide. In 2012, over 1.8 million new cases were reported, with 1.6 million deaths globally (1).

The incidence rates of this disease have increased dramatically in developing countries. Currently around half of lung cancer cases occur in developing countries, particularly in the Arab world where tobacco consumption had increased among both sexes (2).

Individual cigarette smoking is by far the most common risk factor for lung carcinoma; other risks include passive smoke inhalation, residential radon, occupational exposures, infection and genetic susceptibility (3).

Lung cancer has been classified into two clinically important groups: small cell lung cancer (SCLC) and non-small cell lung cancer (NSCLC). NSCLC represents 75–85% of lung cancer, and consists of several types, of which adenocarcinoma (AC) is the most frequent subtype (4).

When possible, differential diagnosis between adenocarcinoma and squamous cell carcinoma is beneficial because targetable driver genetic alterations are mostly identified in adenocarcinoma, and inappropriate drugs need to be avoided for patients with Squamous Cell Carcinoma. Before the 2015 WHO classification, the definitions of adenocarcinoma and Squamous Cell Carcinoma were based on their morphological features with or without mucin staining. Adenocarcinoma was defined as carcinoma with an acinar/tubular structure or mucin production, whereas SqCC was defined as carcinoma with keratinization or intercellular bridges (5).

In the current classification, a solid carcinoma without glandular structures or mucin production but with immunohistochemical positivity for “adenocarcinoma markers” is diagnosed as an adenocarcinoma. Similarly, a solid carcinoma without keratinization, but with immunohistochemical positivity for “Squamous Cell Carcinoma markers is diagnosed as Squamous Cell Carcinoma. These modifications using immunohistochemical evaluations have markedly minimized the proportion of NSCLC diagnosed as large cell carcinoma (6).

Napsin A (Novel aspartic proteinase of the pepsin family) is an enzyme involved in surfactant protein maturation. In the recent years, napsin A has emerged as a useful and broadly utilized diagnostic marker, which is helpful in identifying origin in carcinomas of unknown etiology (7).

Cytokeratin 5/6 is a monoclonal antibody that is intended for laboratory use in the qualitative identification of cytokeratin 5 and 6 proteins by immunohistochemistry (IHC). Studies have shown that Cytokeratin 5/6

antibody reacts with human epidermis and non-keratinizing epithelium (8).

This study aimed to evaluate the expression of Napsin A and CK5/6 in NSCLC, correlate immunohistochemical results of Napsin A and CK5/6 in differentiating between adenocarcinoma and SCC of the lung. assessment of the expression pattern of Napsin A and CK 5/6 as regards the available clinicopathological data available.

Patients and methods

This retrospective study was carried out on 40 lung cases; included 26 adenocarcinoma cases (Resection) and 14 cases of squamous cell carcinoma (Including 7 resection cases and 7 bronchoscopic biopsy cases) which were received from Maadi military hospital and Kobri el Kobba hospital.

Cases were selected according to availability of archived cases with follow-up data in the period from 2017 to 2022. 10 cases of non-neoplastic and non-granulomatous pulmonary lesions were used as control.

The study was approved by the Ethical Committee of faculty of medicine, Benha University. Serial, 5µm-thick, representative sections of formalin-fixed, paraffin-embedded blocks were prepared. One is stained by Hematoxylin and eosin stain subjected to histopathological examination to re-evaluate nature of the lesion as regard

Inclusion criteria were histopathological type (according to WHO 2015) [Adenocarcinoma (AC), squamous cell carcinoma (SCC)], grading (according to WHO Classification 1973) [Grade (I) well differentiated, grade (II) moderately differentiated and grade (III) poorly differentiated. **(SPSS Inc., Chicago, Illinois, USA)**

Immunohistochemical study:

Paraffin embedded tissue sections of 5µm prepared and mounted on positively charged slides to be stained with Napsin A and CK 5/6 antibodies using Biotin streptavidin immuno-peroxidase technique.

Formalin-fixed paraffin –embedded tissue sections were cut at 5um and mounted on positively charged glass slides (Fischer, U SA). Slides were fixed in the oven for 1 hour. Deparaffinized in warm xylene for 30 min then in two changes of cold xylene 30 min each. The slides were rehydrated in a series of graded alcohols to distilled water. Immersion of slides in a buffer solution (citrate monohydrate pH 6) within coplin jar that was placed in water bath and heating in a microwave (800watts) for 5 min then (700 watts) for 6 min and finally at (500 watts) for 15 minutes, as a target antigen retrieval procedure to unmask the antigenic sites and diminish non-specific staining. Buffers were checked after each cycle to prevent the dryness. Slides were allowed to cool at room temperature then rinsed with

distilled water. Slides were placed in phosphate buffer saline (PBS)(pH 7.6) buffer for 5 minutes. Tissue sections were not allowed to dry out at any point during staining and rehydration procedure. Excess buffer was tapped off. Enough peroxides block (2-3 drops) was applied to cover specimens and incubated for 15 minutes. Then, slides were rinsed gently with buffer solution. Excess buffer was tapped off. Add 2 drops of diluted Napsin A and CK5\6 antibody were added to each section to cover the section completely, slides were incubated in humidity chamber for 30 minutes. Slides were rinsed in distilled water then in PBS. Enough biotinylated antibodies (2-3 drops) were applied to cover the specimen and incubated for 30 minutes. Then Slides were rinsed in distilled water then in PBS. Enough strep avidin peroxidase was applied to cover specimen and incubated for 30 minutes. Slides were rinsed in distilled water.

Substrate-chromogen:

Enough of freshly prepared DAB+ substrate-chromogen solution (2 drops of DAB and 1 ml of DAB substrate then mix gently)was applied to cover specimen and incubated for 5minutes until color intensity had been reached. Slides then rinsed gently with distilled water from a wash bottle. Slides were immersed in a bath of Mayer's hemoatoxylin for 1 minute depending on the strength of hematoxylin. Slides then rinsed gently in distilled water bath, dehydrated in 95% ethyl alcohol, and followed by absolute ethanol. Then slides were cleared in xylene, using two changes. Cover slips were mounted using two drops of DPX mounting medium (7,8,9).

Each staining session included both positive and negative control slides:

Negative control: Negative control slides were processed in the same immunostaining procedure in omission of the primary antibody.

Positive control: Sections from normal lung tissue (pneumocytes type II) as internal control was served as positive control for Napsin A. Sections from skin were served as positive control slides for CK 5/6.

Interpretation of Immunostaining Slides: Examination of negative and positive control slides was preformed first to rule out nonspecific staining and to judge the effectiveness of the technique, and the reagents respectively. Examination and assessment of Napsin A and CK 5/6 stained non-small cell lung carcinomas slides for: The pattern of Napsin A and Ck 5/6 immunoreactivity: Napsin A was cytoplasmic/ membranous according to CK 5/6 was cytoplasmic stained according to (8,9).

Scoring of Napsin A immunoreactivity: In order to evaluate Napsin A immunostaining, tumour cells are considered positive when showing cytoplasmic and/or membranous

staining. 5% of positive neoplastic cells were used as cut-off point.

Immunoreactivity of Napsin A was scored based on percentage of expression as follows: Low Napsin A expression: staining in 5-10% of tumor cells. Moderate expression: staining in 11-50% of tumor cells. Extensive expression: staining in >50% tumor cells. (10,11)

Scoring of CK 5/6 immunoreactivity: To evaluate CK5/6 immunostaining, Neoplastic cells are considered positive when showing cytoplasmic staining. 5% of positive neoplastic cells were used as cut-off point Immunoreactivity of CK5/6 was scored based on percentage of expression as follows: Negative: no staining. Focal expression; staining in 5% to 10% of neoplastic cells. Patchy expression; staining in 11% to 50% of Neoplastic cells. Diffuse expression; staining in >50% of neoplastic cells (12). (SPSS Inc., Chicago, Illinois, USA)

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Statistical analysis

The data were coded, entered and processed on a personal computer using statistical program for social science (SPSS) (version 16). Qualitative data was summarized using frequency and percentages. Association between different clinicopathological variables as well as both of Napsin A and CK5\6 expression was determined using the Pearson correlation coefficient. The differences were assessed by determining the probability (P value) which was considered as statistically significance when p value < 0.05. If p value <0.01, it was considered as highly statistical significance and considered as statistically insignificant when p value > 0.05.

Results

Table 1 shows Socio-demographic data, smoking status, histopathological type of tumor, tumor grade distribution, regional lymph node metastasis and tumor site distribution among the studied 40 cases and histological subtypes of studied adenocarcinoma biopsy specimens.

Table 2 shows Tumor grades, T stage, Lymph Node involvement, M stage, and TNM Stage of studied adenocarcinoma cases.

This comparative study between the 2 groups revealed non-significant difference as regards age, sex, smoking status of the patients ($p > 0.05$). Significant high LN involvement in squamous cell carcinoma compared to adenocarcinoma group ($p < 0.05$). While there was non-significant difference between the 2 groups as regards all the remaining pathological data (tumor data) ($p > 0.05$). **Table 3**

Highly significant increase in Napsin A score and expression, in ADC group (All positive Napsin A cases were ADC); compared to SSC group ($p < 0.001$). While there was highly significant increase in CK5/6 score and expression,

in SSC group (All positive CK5/6 cases were SSC); compared to ADC group ($p < 0.00$). **Table 4**

Pearson's correlation analysis showed that ADC had a highly significant positive correlation with Napsin A score ($p < 0.01$). Pearson's correlation analysis showed that SSC and LN had a highly significant positive correlation with CK5/6 score ($p < 0.01$). LN had significant positive correlation with CK5/6 score ($p < 0.05$). **Table 5**

Logistic regression analysis showed that; after applying (Forward method) and entering some predictor variables; the decrease in LN; had an independent effect on increasing the probability of Napsin A expression; with significant statistical difference ($p = 0.043$). Logistic regression analysis shows that; after applying (Forward method) and entering some predictor variables; the increase in SSC histology had an independent effect on increasing the probability of CK5/6 expression; with significant statistical difference ($p = 0.0004$). **Table 6**

By using ROC-curve analysis, Napsin A score predicted ADC patients from SSC patients, with perfect (100%) accuracy, sensitivity= 100% and specificity= 100% ($p < 0.01$). By using ROC-curve analysis, CK5/6 score predicted SSC patients from ADC patients, with excellent (92%) accuracy, sensitivity= 97% and specificity= 85% ($p < 0.01$). **Table 7**

Table 1: Socio-demographic data, smoking status, histopathological type of tumor, tumor grade distribution, regional lymph node metastasis and tumor site distribution among the studied 40 cases and histological subtypes of studied adenocarcinoma biopsy specimens

Variables		**Frequency (%) / Mean ± SD**
Age (years)		64.5 ± 4.4
Gender	Female	6 (15%)
	Male	34 (85%)
	Total	40 (100%)
Smoking status	Non-Smoker	4 (10%)
	Current Smoker	36 (90%)
Histopathological type of tumor	Adenocarcinoma	26(65%)
	Squamous cell carcinoma	14(35%)
Tumor Grade	Well differentiated (GI)	3(7.5%)
	Moderately differentiated (GII)	25(62.5%)
	Poorly differentiated (GIII)	12(30%)
Regional lymph node metastasis		10(30.3%)
Tumor site	Right	24(60%)
	Left	16(40%)
Histological Subtypes	Solid	11(42.3%)
	Acinar	6(23.1%)
	Papillary	6(23.1%)
	Mucinous	3(11.5%)

Table 2*:* Tumor grades, T stage, Lymph Node involvement, M stage, and TNM Stage of studied adenocarcinoma cases

Grade	Number of Cases	Percentage
Well Differentiated (GI)	2	7.7
Moderately Differentiated (GII)	16	61.5
Poorly Differentiated (GIII)	8	30.8
T stage		
T1	8	28
T2	18	72
T3	0	0
T4	0	0
Lymph Node		
Positive (+ve)	5	19.2
Negative (-ve)	21	80.8
M stage		
M0	23	88.5
M1	3	11.5
TNM Stage		
Stage I	4	15.8
Stage II	17	65.2
Stage III	2	7.6
Stage IV	3	11.4

Table 3: Comparison between the 2 groups as regards Socio-demographic data, Smoking status, Biopsy data (tumor data):

Variable		Adenocarcinoma "ADC group (26)	Squamous cell carcinoma "SSC group (14)	Student's *t* test
		Mean ± SD	**Mean ± SD**	**P value**
Age (years)		64.7 ± 4.2	63.4 ± 5.38	= 0.480
Gender	**Female**	4 (15.4%)	2 (14.3%)	= 0.2743
	Male	22 (84.6%)	12 (85.7%)	
Non-Smoker (4)		3 (11.5%)	1 (7.1%)	= 0.6983
Current Smoker (36)		23 (88.5%)	13 (92.9%)	= 0.6812
Size of tumor **(cm)**		3.5 ± 0.88	3.1 ± 0.74	= 0.352
Histopathologic al pattern of tumor	**Solid**	11 (42.3%)		= 0.8542
	Acinar	6 (23.1%)		
	Papillary	6 (23.1%)		
	Mucinous	3 (11.5%)		
	Non Keratinizing		6 (42.9%)	
	Keratinizing		4 (28.6%)	
	Baseloid		4 (28.6%)	
Grade of tumor	**Well differentiated**	2 (7.7%)	2 (14.3%)	= 0.5149
	Moderately differentiated	16 (61.5%)	10 (71.4%)	
	Poorly differentiated	8 (30.8%)	2 (14.3%)	
LN	**+ve**	5 (19.2%)	8 (57.1%)	=0.032*

* Percentage of Column Total.

Table 4: Immunohistochemical staining among 40 biopsy specimens

Variables			Frequency (%) / Mean ± SD	
Napsin A score (0 - 3)			1.7 ± 1	
Napsin A		Expressed	24 (60%)	
CK5/6 score (0 - 3)			0.27 ± 0.64	
CK5/6		Expressed	11 (27.5%)	
		ADC group (26)	**SSC group (14)**	**P value**
Napsin A score (0 - 3)		2 ± 0.65	0 ± 0	< 0.001*
CK5/6 score (0 - 3)		0.03 ± 0.17	1.42 ± 0.78	< 0.001*
Napsin A	Expressed	24 (92.3%)	0 (0%)	< 0.001*
CK5/6	Expressed	0 (0%)	11 (78.0%)	< 0.001*

*: Significant P value

Table 5: Pearson's correlation analysis for clinical / pathological data / tumor data Factors associated with Immunohistochemical Napsin A score and Immunohistochemical CK5/6 score

Associated Factor	Napsin A score	
	r	**P value**
Age **(years)**	0.0134	=0.9346
Gender	0.00776	= 0.930
Non-Smoker	0.126	=0.4382
Current Smoker	-0.0347	=0.8317
Ex-Smoker	0.0370	=0.8205
Size of tumor **(cm)**	0.0192	=0.9064
Histopathological type (ADC)	0.9509	< 0.001**
Histopathological pattern of tumor	0.114	= 0.952
Tumor grade	0.118	=0.4692
LN	-0.195	=0.2279
Associated Factor	**CK5/6 score**	
	r	**P value**
Age **(years)**	-0.114	=0.4824
Gender	0.0572	= 0.812
Non-Smoker	-0.0412	=0.8007
Current Smoker	-0.0819	=0.6155
Ex-Smoker	-0.0734	=0.6526
Size of tumor **(cm)**	-0.279	=0.0817
Histopathological examination	0.9160	< 0.001**
Histopathological pattern of tumor	0.0368	= 0.777
Grade of tumor	-0.0665	=0.6833
LN	0.359	=0.022*

r: Pearson's rho (correlation coefficient).

Table 6: Logistic regression model for the Factors affecting Napsin A and CK5/6 expression using forward method:

Predictor Factor	Coefficient	OR	P value
(Constant)	2.19722		
LN	-1.79176	0.1667	0.043*
Predictor Factor	Coefficient	OR	P value
(Constant)	-3.46574		
SSC histology	5.25750	192.0000	0.0004**

Other factors excluded from the model as (p value > 0.1). OR: odds ratio.

Table 7: ROC -curve of Immunohistochemical staining to predict patients with ADC from patients with SSC:

Variable	AUC	Best Cut off point (Criterion)	Sensitivity (%)	Specificity (%)	P value
Napsin A score	1.000	>0	100	100	<0.0001**
CK5/6 score	0.922	≤0	96.97	85.71	<0.0001**

ROC (Receiver operating characteristic), AUC= Area under curve, SE= Standard Error.

Discussion

In the current study the mean age of all patients was (64.5 ± 4.4) years. Regarding gender of the patients, the majority (85%) of patients was males, while (15%) were females. Regarding smoking status, (7.5%) of patients were non-smokers, (90%) were current smokers, and (2.5%) had passive smoker.

Include 53 patients ranged in age from 47 to 87 years (mean 67.3, median 68). Twenty-seven patients were females, and 26 patients were males (9).

In the current study regarding biopsy data (tumor data); the average size of tumor was (3.4 ± 0.8) cm, with (25%) having LN metastasis.

Similarly found that the size of the tumors ranged from 1.0 to 9.8 cm (mean 3.1 cm, median 2.8 cm) (9).

Studying 60 patients who presented with solitary lung lesions. The age of the patients ranged from 47 to 79 with a median of 46, and a mean of 64.13. Sixteen patients were females representing 26.7% of patients, while 44 were males representing 73.3% of patients. The right lung showed 63.3% of lesions, while the remaining 36.7% of lesions was in the left lung. Twenty percent of lung masses was <2 cm, 65% of masses ranged between 2 and 5 cm, while 15% was >5 cm (10).

In the current study Regarding Histopathological examination, (65%) of patients had Adenocarcinoma, and (35%) had Squamous cell carcinoma.

In contrast to found that a cohort of 43 malignant cases consisting of ADC (10 cases), SCC (15 cases) (11).

In the current study regarding the histopathological pattern of tumor; (15%) of patients had Acinar and Mucinous pattern, (7.5%) had Papillary pattern, and (62.5%) had Solid pattern. Regarding Grade of tumor, (7.5%) of patients had Well differentiated, (62.5%) had Moderately differentiated, and (30%) had Poorly differentiated.

In a study by found that by histologic examination, the ADCs revealed variable histologic features consisting of acinar, lepidic, papillary, solid, and micropapillary pattern. The SCCs showed a wide range of differentiation from well to poor degree (12).

In the current study Regarding Immunohistochemical staining; the

average Napsin A score was (1.7 ± 1), with (82.5%) had Napsin A expression, and the average CK5/6 score was (0.27 ± 0.64), with (17.5%) had CK5/6 expression.

In the current study Comparative study between the 2 groups revealed non-significant difference as regards age, sex and smoking of the patients ($p > 0.05$).

On the other hand, Singh and co-workers found that among the 520 NSCLC patients, mean [standard deviation (SD)] age in groups I, II and III was 54.5 (12.5), 58.6 (9.9) and 61.2 (9.4) years respectively ($P<0.001$). Percentage of males in the three groups was 48.1%, 88.0%, and 97.9% ($P<0.001$) this may be due to the comparison between the three groups of NSCLC (13).

In the current study Comparative study between the 2 groups revealed; highly significant decrease in LN involvement in ADC group; compared to SSC group ($p < 0.05$).

In study patients with adenocarcinoma, smokers never showed a younger age at diagnosis (54.2 ± 12.7vs. 59.3 ± 9.4, p adjusted<0.001), a lower risk for lymph node metastasis than smokers (7,6% vs. 19.5%, p adjusted<0.001) and less severe disease as indicated by lower percentages of patients with TNM stage of III or IV (5.5% vs. 14.7%, p adjusted<0.001). By contrast, these associations were not observed in 50 patients with squamous cell carcinoma (14).

In the current study Comparative study between the 2 groups revealed; highly significant increase in Napsin A score and expression, in ADC group; compared to SSC group ($p < 0.001$).

Study found that in biopsy-stained slides 27 (58.7%) tumors exhibited positive Napsin A expression. All ADC cases (13), 4 (14.8%) SCC (showed granular cytoplasmic reactivity) and 10 (58.8%) LCC were positive for Napsin A expression (15)

In the current study comparative study between the 2 groups revealed; highly significant increase in CK5/6 score and expression, in SSC group; compared to ADC group ($p < 0.001$).

Similarly found that the results showed that the SCC cases were all positive for CK5/6 (100%) with a cytoplasmic stain pattern (Fig. C-1) and ADC cases were rarely positive (10%), significantly different by statistical analysis (11).

In the current study comparative study between the 2 groups revealed; highly significant increase in CK5/6 score and expression, in SSC group; compared to ADC group ($p < 0.001$).

In agreement with our study found that Most of SCC cases were positive for CK5/6 (90%), and CK5/6 was almost negative in ADC (12).

In the current study Pearson's correlation analysis shows that ADC had a highly significant positive correlation with Napsin A score ($p < 0.01$).

In agreement with our result found that there was significant statistical correlation of Napsin-A expression in ADC (16).

In the current study Pearson's correlation analysis shows that SSC had a highly significant positive correlation with CK5/6 score ($p < 0.01$ respectively).

In consistent with our result found that Univariate analysis showed that both high levels of p63 and CK5/6 expression and p63+CK5/6 co-expression correlated with the prognosis of N0 lung SQCC patients. In addition, the co-expression of p63+CK5/6 perfectly correlated with the expression of p63 and CK5/6 ($rs = 1$). Therefore, the expression of p63 and CK5/6 and the co-expression of p63+CK5/6 were considered prognostic factors and separately included in the Cox regression analysis. Other included factors were age, gender, smoking history, (pT) stage, and degree of differentiation (17).

In the current study logistic regression analysis shows that; after applying (Forward method) and entering some predictor variables; the decrease in LN had an independent effect on increasing the probability of Napsin A expression; with significant statistical difference ($p = 0.043$). Like as they found that Napsin A expression showed significant correlation to lymphatic metastasis (18).

In the current study logistic regression analysis shows that; after applying (Forward method) and entering some predictor variables; the increase in SSC histology; had an independent effect on increasing the probability of CK5/6 expression; with significant statistical difference ($p = 0.0004$).

This was explained as presence of CK5/6 expression was more frequent in endometrioid tumors with squamous differentiation, while loss of CK5/6 expression (54%) was significantly associated with high FIGO stage, reduced beta-catenin expression, MSI and reduced patient survival ($p = 0.0001$); the latter was also found within the endometrioid subgroup ($p = 0.0004$). Multivariate survival analysis revealed that loss of CK5/6 expression had an independent prognostic impact in addition to well-known prognostic variables. Expression of both markers increased in simple hyperplasia compared with normal endometrium. In complex hyperplasia, p63 expression was also increased, whereas CK5/6 was positive in areas with squamous differentiation only (19).

In the current study by using ROC-curve analysis, Napsin A score predicted patients with ADC from patients with SSC, with perfect (100%) accuracy, sensitivity= 100% and specificity= 100% ($p < 0.01$). In contrast to other studies found that Napsin A The sensitivity, specificity, positive predictive value, and negative predictive value of the markers for AC and SCC were 64 (16/25), 100 (23/23), 100 (16/16), 72 (23/32) respectively (20).

Conclusion

From our findings we can conclude that the score on Napsin A can predict patients with ADC from patients with SSC. The increase in SSC histology had an independent effect on increasing the probability of CK5/6 expression. The decrease in LN had an independent effect on increasing the probability of Napsin A expression.

References

1. Siegel RL, Miller KD, Jemal A. Cancer statistics, 2015. CA: a cancer journal for clinicians. 2015;65.
2. Temraz S, Charafeddine M, Mukherji D, Shamseddine A. Trends in lung cancer incidence in Lebanon by gender and histological type over the period 2005-2008. J Epidemiol Glob Health. 2017;7:161-7.
3. de Groot PM, Wu CC, Carter BW, Munden RF. The epidemiology of lung cancer. Translational Lung Cancer Research. 2018;7:220-33.
4. Mohammed F, Pity I, Safo A, Izac A. Cytokeratin 5/6, p63 and TTF-1 Immunomarker Use in Tiny Non-small Cell Lung Cancer. Journal of Cancer Prevention & Current Research. 2018;9:191-5.
5. Costa GJ, de Mello MJG, Ferreira CG, Bergmann A, Thuler LCS. Increased incidence, morbidity and mortality rates for lung cancer in women in Brazil between 2000 and 2014: An analysis of three types of sources of secondary data. Lung Cancer. 2018;125:77-85.
6. Travis WD. WHO classification of tumors. Pathology and Genetics of Tumors of the Lung, Pleura, Thymus and Heart. 2004:128-36.
7. Inamura K. Lung Cancer: Understanding Its Molecular Pathology and the 2015 WHO Classification. Front Oncol. 2017;7:193.
8. Nishino M, Hoang MP, Della Pelle P, Morales-Oyarvide V, Huynh TG, Mark EJ, et al. Napsin A/p40 antibody cocktail for subtyping non-small cell lung carcinoma on cytology and small biopsy specimens. Cancer Cytopathol. 2016;124:472-84.
9. Khayyata S, Yun S, Pasha T, Jian B, McGrath C, Yu G, et al. Value of P63 and CK5/6 in distinguishing squamous cell carcinoma from adenocarcinoma in lung fine-needle aspiration specimens. Diagn Cytopathol. 2009;37:178-83.
10. Ezzat Nel S, Tahoun N. The role of Napsin-A and Desmocollin-3 in classifying poorly differentiating non-small cell lung carcinoma. J Egypt Natl Canc Inst. 2016;28:13-22.
11. Pu RT, Pang Y, Michael CW. Utility of WT-1, p63, MOC31, mesothelin, and cytokeratin (K903 and CK5/6) immunostains in differentiating adenocarcinoma, squamous cell carcinoma, and malignant mesothelioma in effusions. Diagn Cytopathol. 2008;36:20-5.
12. Kim MJ, Shin HC, Shin KC, Ro JY. Best immunohistochemical panel in distinguishing adenocarcinoma from squamous cell carcinoma of lung: tissue microarray assay in resected lung cancer specimens. Ann Diagn Pathol. 2013;17:85-90.
13. Singh N, Aggarwal AN, Gupta D, Behera D, Jindal SK. Quantified smoking status and non-small cell lung cancer stage at presentation: analysis of a North Indian cohort and a systematic review of literature. J Thorac Dis. 2012;4:474-84.
14. Shan L, Zhang L, Zhu X, Wang Z, Fang S, Lin J, et al. Chinese never smokers with adenocarcinoma of the lung are younger and have fewer lymph node metastases than smokers. Respir Res. 2022;23:293.
15. Samir RM, Osman GS, El-Deftar MMF. Diagnostic Value of Napsin A in Small Biopsy of Non-Small Cell Lung Carcinoma. The Egyptian Journal of Hospital Medicine. 2021;83:1297-301.
16. Sharma T, Das P, Panigrahi R, Rao CM, Rath J. Immunocytochemical Evaluation of TTF-1, Napsin-A, and p-63 for Subtyping of Non-Small Cell Lung Carcinoma and Clinicopathological Correlation. J Cytol. 2022;39:180-7.
17. Ma Y, Fan M, Dai L, Kang X, Liu Y, Sun Y, et al. Expression of p63 and CK5/6 in early-stage lung squamous cell carcinoma is not only an early diagnostic indicator but also correlates with a good prognosis. Thorac Cancer. 2015;6:288-95.
18. Wei YH. Effect of Napsin A and Circulating Tumor Cells with Mesenchymal Phenotype in Lung Adenocarcinoma. 2021.

19. Stefansson IM, Salvesen HB, Akslen LA. Loss of p63 and cytokeratin 5/6 expression is associated with more aggressive tumors in endometrial carcinoma patients. Int J Cancer. 2006;118:1227-33.

20. Zhao W, Wang H, Peng Y, Tian B, Peng L, Zhang DC. ΔNp63, CK5/6, TTF-1 and napsin A, a reliable panel to subtype non-small cell lung cancer in biopsy specimens. Int J Clin Exp Pathol. 2014;7:4247-53.

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